

#2920 Store at -20°C

Akt (pan) (40D4) Mouse mAb

100 µl
 (20 western blots)

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This product is intended for research purposes only. This product is not intended to be used for therapeutic or diagnostic purposes in humans or animals.

Applications	Species Cross-Reactivity*	Molecular Wt.	Isotype
W, IP, IHC-P, IF-IC, F Endogenous	H, M, R, Mk	60 kDa	Mouse IgG1κ**

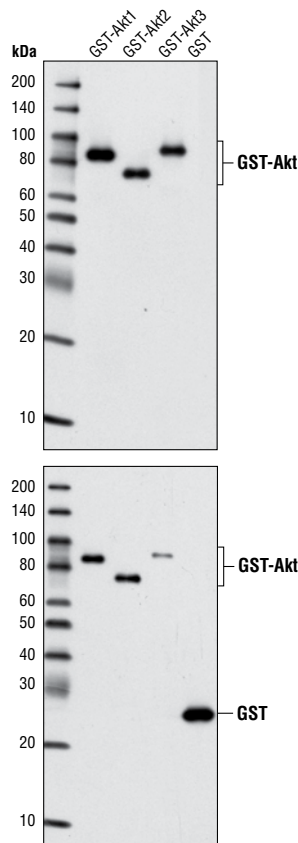
Background: Akt, also referred to as PKB or Rac, plays a critical role in controlling survival and apoptosis (1-3). This protein kinase is activated by insulin and various growth and survival factors to function in a wortmannin-sensitive pathway involving PI3 kinase (2,3). Akt is activated by phospholipid binding and activation loop phosphorylation at Thr308 by PDK1 (4) and by phosphorylation within the carboxy terminus at Ser473. The previously elusive PDK2 responsible for phosphorylation of Akt at Ser473 has been identified as mammalian target of rapamycin (mTOR) in a rapamycin-insensitive complex with rictor and Sin1 (5,6). Akt promotes cell survival by inhibiting apoptosis by phosphorylating and inactivating several targets, including Bad (7), forkhead transcription factors (8), c-Raf (9) and caspase-9. PTEN phosphatase is a major negative regulator of the PI3 kinase/Akt signaling pathway (10). LY294002 is a specific PI3 kinase inhibitor (11).

Another essential Akt function is the regulation of glycogen synthesis through phosphorylation and inactivation of GSK-3α and β (12,13). Akt may also play a role in insulin stimulation of glucose transport (12).

In addition to its role in survival and glycogen synthesis, Akt is involved in cell cycle regulation by preventing GSK-3β mediated phosphorylation and degradation of cyclin D1 (14) and by negatively regulating the cyclin dependent kinase inhibitors p27 Kip (15) and p21 Waf1/CIP1 (16). Akt also plays a critical role in cell growth by directly phosphorylating mTOR in a rapamycin-sensitive complex containing raptor (17). More importantly, Akt phosphorylates and inactivates tuberlin (TSC2), an inhibitor of mTOR within the mTOR-raptor complex (18). Inhibition of mTOR stops the protein synthesis machinery due to inactivation of its effector, p70 S6 kinase and activation of the eukaryotic initiation factor 4E binding protein 1 (4E-EP1), an inhibitor of translation (18,19).

Specificity/Sensitivity: Akt (pan) (40D4) Mouse mAb detects endogenous levels of total Akt protein. This antibody does not cross-react with other related proteins.

Source/Purification: Monoclonal antibody is produced by immunizing animals with a synthetic peptide at the carboxy-terminal sequence of human Akt.



Western blot analysis of recombinant Akt1, Akt2, Akt3 and GST proteins using Akt (pan) (40D4) Mouse mAb (upper) and GST (91G1) Rabbit mAb #2625 (lower).

Entrez-Gene ID #207
Swiss-Prot Acc. #P31749

Storage: Supplied in 10 mM sodium HEPES (pH 7.5), 150 mM NaCl, 100 µg/ml BSA, 50% glycerol and less than 0.02% sodium azide. Store at -20°C. Do not aliquot the antibody.

*Species cross-reactivity is determined by western blot.

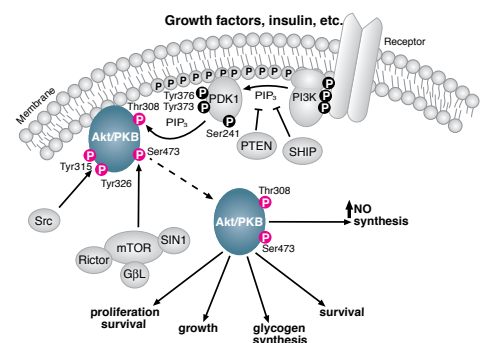
**Anti-mouse secondary antibodies must be used to detect this antibody.

Recommended Antibody Dilutions:

Western blotting	1:2000
Immunoprecipitation	1:50
Immunohistochemistry (Paraffin)	1:250
Unmasking buffer:	Citrate
Antibody diluent:	SignalStain® Antibody Diluent #8112
Immunofluorescence (IF-IC)	1:50
Flow Cytometry	1:50

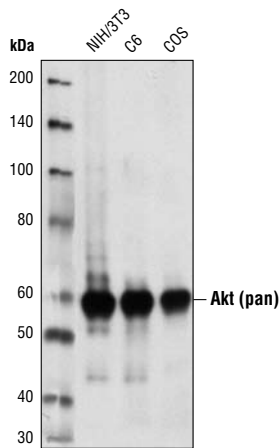
For application specific protocols please see the web page for this product at www.cellsignal.com.

Please visit www.cellsignal.com for a complete listing of recommended companion products.

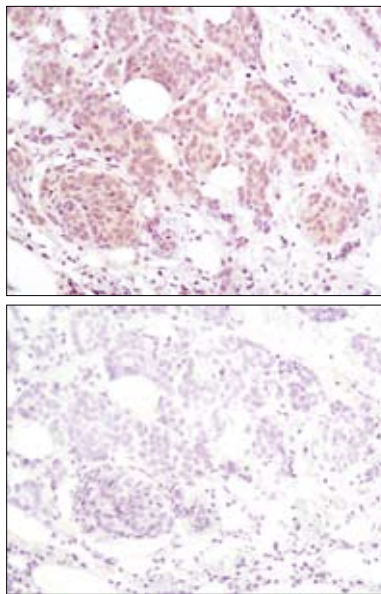


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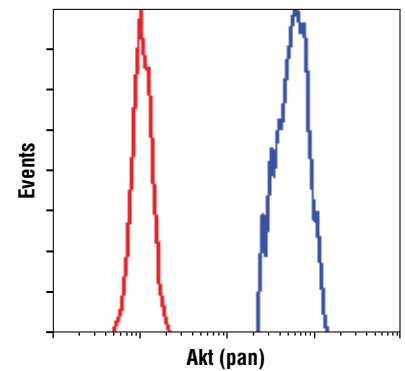
Applications Key: W—Western IP—Immunoprecipitation IHC—Immunohistochemistry ChIP—Chromatin Immunoprecipitation IF—Immunofluorescence F—Flow cytometry E-P—ELISA-Peptide
Species Cross-Reactivity Key: H—human M—mouse R—rat Hm—hamster Mk—monkey Mi—mink C—chicken Dm—D. melanogaster X—Xenopus Z—zebrafish B—bovine
 Dg—dog Pg—pig Sc—S. cerevisiae Ce—C. elegans Hr—Horse All—all species expected Species enclosed in parentheses are predicted to react based on 100% homology.



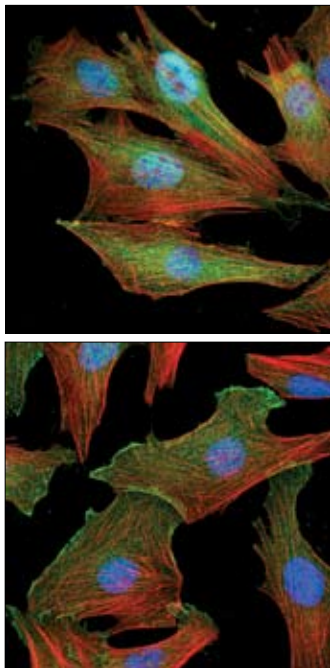
Western blot analysis of extracts from NIH/3T3, C6 and COS cells using Akt (pan) (40D4) Mouse mAb.



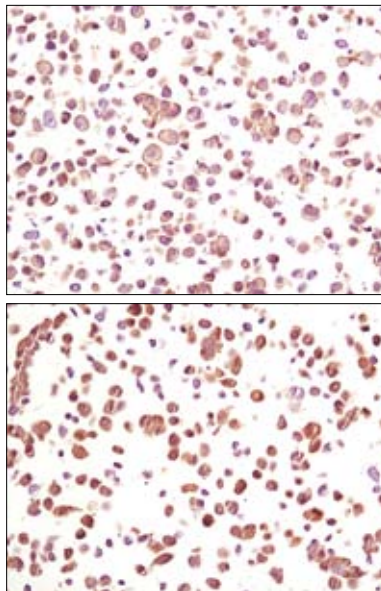
Immunohistochemical analysis of paraffin-embedded human breast carcinoma using Akt (pan) (40D4) Mouse mAb in the presence of control peptide (upper) or Akt (pan) Blocking Peptide #1085 (lower).



Flow cytometric analysis of untreated Jurkat cells using Akt (pan) (40D4) Mouse mAb (blue) compared to a nonspecific negative control antibody (red).



Confocal immunofluorescent analysis of C2C12 cells, either LY294002-treated (upper) or insulin-treated (lower), using pan-Akt (40D4) Mouse mAb (green). Actin filaments have been labeled with DY-554 phalloidin (red). Blue pseudo-color = DRAQ5[®] #4084 (fluorescent DNA dye).



Immunohistochemical analysis using Akt (pan) (40D4) Mouse mAb on SignalSlide™ Phospho-Akt (Ser473) IHC Controls #8101 (paraffin-embedded LNCaP cells, untreated (upper) or LY294002-treated (lower)).

Background References:

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